

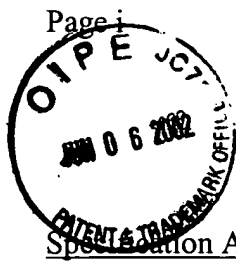
REMARKS

Applicants, in compliance with 37 C.F.R. 1.821 (f), hereby state that the information recorded in computer readable form submitted herewith is identical to the written sequence listing. Applications, in compliance with 37 C.F.R. 1.821 (g), further state that no new matter is added.

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Respectfully submitted,

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MARKED-UP VERSION OF AMENDMENTS:Specification Amendments Under 37 C.F.R. § 1.121(b)(1)(iii)

Please replace the paragraph at page 56, lines 3 through 20 with the paragraph below, which is marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph:

Example 1. Detection Of Nucleotide At Predetermined Position Using Probe Complementary To The Primer

Detection of SNPs was also performed by FRET minisequencing using a probe which is fully complementary to the primer. The primer pBA was designed to anneal to pBluescript (A562C) so that the dideoxynucleotide to be incorporated is a ddCTP.

pBA 5'- GGATGTGCTGCAAGGCGATT -3' (SEQ ID NO: 1)

pAntiBA 3'- (P)CCTACACGACGTTCCGCTAA (F) -5' (SEQ ID NO: 2)

pBA and pAntiBA were synthesized and HPLC purified by Genset Corporations (La Jolla, CA). pAntiBA20 was labeled with Fluorecein at 5'-end, and blocked with a phosphate group at its 3'-end. The relative orientation of the primers (above) are arranged to facilitate viewing of how they will hybridize to each other. 25 µl reactions contained 200 nM ROX-ddC, 4 U polymerase, 250 nM pBA 250 nM pAntiBA, and 200 nM pBluescript in 1 x polymerase reaction buffer. Negative control lacked DNA template (pBluescript). Thermal cycling was performed in the Applied Biosystems Prism 7700 Sequence Detector. Thermal cycling conditions were performed by initial denaturing step at 95°C for 2 minutes, followed by 30 cycles at 95°C for 30 s, 50°C for 1 min, and 57°C for 30 s. The fluorescent intensities were acquired during the annealing/extension phase of the primer extension cycles. The analysis was done using the multicomponent data from the Applied Biosystems 7700 Sequence Detector. Figure 4 illustrates that the positive control (A4 well) shows a ROX signal increase due to FRET from Fluorescein compared to the negative control (A3 well).

Please replace the paragraph at page 56, lines 22 through 26, to page 57, lines 1 through 17 with the paragraph below, which is marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph:

Example 2. Detection Of Nucleotide At Predetermined Position Using a probe partially complementary to the primer

Detection of SNPs was also performed by FRET minisequencing using a probe which is partially complementary to the primer. The primer pJ was designed to anneal to pBluescript (A562C) so that the dideoxynucleotide to be incorporated is a ddCTP.

pJ 5'- GAGGCTCGGAGCGGTTAAACGGATGTGCTGCAAGGCGATT -3' (SEQ ID NO: 3)

Non-complementary to
template
Complementary to
template

pAntiJ 3'- (P)CTCCGAGCCTCGCCAATTTG(F) -5' (SEQ ID NO: 4)

Oligos were synthesized and HPLC purified by Genset Corporations (La Jolla, CA). pAntiJ was labeled with Fluorecein at 5'-end, and blocked with a phosphate group at its 3'-end. The relative orientation of the primers (above) are arranged to facilitate viewing of how they will hybridize to each other.

25 µl reactions contained 200 nM ROX-ddC, 4 U polymerase, 250 nM pJ 250 nM pAntiJ, and 200 nM pBluescript in 1 x polymerase reaction buffer. Negative control lacked DNA template (pBluescript). Thermal cycling was performed in the Applied Biosystems Prism 7700 Sequence Detector. Thermal cycling conditions were performed by initial denaturing step at 95°C for 2 minutes, followed by 30 cycles at 95°C for 30 s, 50°C for 1 min, and 57°C for 30 s. The fluorescent intensities were acquired during the annealing/extension phase of the primer extension cycles. The analysis was done using the multicomponent data from the Applied Biosystems 7700 Sequence Detector. Figure 5 illustrates that the positive control (B2 well) shows a ROX signal increase due to FRET from Fluorescein compared to the negative control (B1 well).

Please replace the paragraph at page 57, lines 18 through 25, with the paragraph below, which is marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph:

Example [4] 3.Detection Of Nucleotide At Predetermined Position Using a quencher molecule and a probe partially complementary to the primer

Detection of SNPs was also performed by FRET minisequencing using a probe containing a quencher (Figure 6). The primer pJ was designed to anneal to pBluescript (A562C) so that the dideoxynucleotide to be incorporated is a ddCTP.

pJ 5'- GAGGCTCGGAGCGGTAAACGGATGTGCTGCAAGGCGATT -3' (SEQ ID NO:3)

Non-complementary to template Complementary to template

pAntiJ-BHQ 3'- (P)CTCCGAGCCTCGCCAATTTG(BHQ2) -5' (SEQ ID NO. 4)